



## EFFECT OF RINGER'S SOLUTION ON TAIL-REGENERATION IN THE TADPOLE OF *RANA PALUSTRIS*

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It has been shown that the regenerative activity of the fresh-water oligochaete *Lumbriculus* is decreased in balanced salt solutions (Ringer) which are isotonic, or nearly so, with the body fluids of that worm (Sayles, 1928). Also, in the case of certain marine invertebrates, it has been found that dilutions of the sea water increase the rate of regeneration (Loeb, 1892; Goldfarb, 1914; Lloyd, 1914; Keil, 1932 and others). In the case of marine forms, of course, the body fluids are approximately isotonic with undiluted sea-water.

With these facts in mind a study was made to determine whether or not a similar slowing-up of regeneration in isotonic salt solutions might occur in the case of frog tadpoles.

Tadpoles of *Rana palustris*, varying in total length from 40 to 65 millimeters and with small posterior legs, were used. They were first anesthetized in a solution of approximately 1 part of chloretone in 2,200 of water. Either 15 or 20 mm. of the tail were then removed.

The first experiments on this material were carried out during the summer of 1927. Then, in 1929 and again in 1932, results essentially the same as those of the first year were obtained.

The results of two experiments, typical of a number performed, may be presented to indicate the effect of Ringer solutions on the length and form of the bud of new tissue. In one instance, four tadpoles were cut. Tadpole *A* (52 mm. total length) was placed in water. *B* (55 mm. long), *C* (50 mm. long), and *D* (40 mm. long) were placed in buffered frog Ringer. All four had 15 mm. of tail removed. After two weeks the lengths of the new buds were as follows: *A*, 9.0 mm.; *B*, 1.9 mm.; *C*, 3.6 mm.; *D*, 3.4 mm. All of these were killed at the end of the third week, at which time no marked change in the relative lengths of the buds had occurred.

In another experiment four tadpoles, each 40 mm. in total length, had 20 mm. removed from the tail. *A* was placed in tap-water; *B* in Ringer of 0.5 usual strength; *C* in ordinary frog Ringer; and *D* in Ringer of 1.1 usual strength. After twenty-three days the lengths of

the buds were as follows: *A*, 12.3 mm.; *B*, 9.3 mm.; *C*, 7.1 mm.; *D*, 6.4 mm.

In all individuals regenerating in tap-water the tail bud is complete with well-formed swimming membrane. Tadpoles regenerating in 0.5 Ringer usually produce complete tails of the ordinary type. In some cases, however, the musculo-skeletal axis is complete but the caudal membrane is much reduced or even absent in part of the bud. A regenerate of this type is shown in Fig. 1.

In straight frog Ringer about two-thirds of the short buds have well-developed membranes. These apparently differ from the control animals only in the length of the bud. In two of these tadpoles, however, the buds formed have only narrow, somewhat irregular portions of the membrane (Fig. 2). In the cases of the other tadpoles regenerating in this solution, however, a slender bud is formed with little or no caudal membrane. Figure 3 shows such a regenerate. This particular

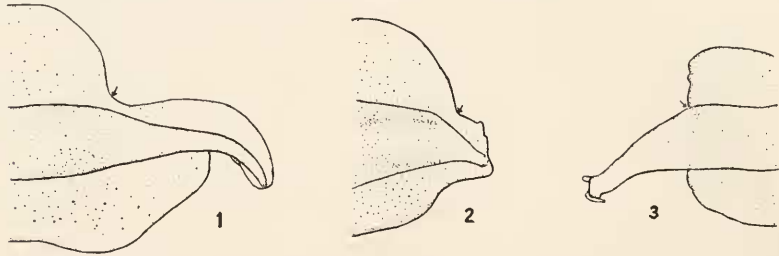


FIG. 1. A tail bud formed after 11 days by a tadpole regenerating in 0.5 Ringer solution.  $\times 4$ .

FIG. 2. A tail bud formed after 29 days of regeneration in straight Ringer solution.  $\times 3$ .

FIG. 3. Slender bud consisting merely of musculo-skeletal axis with two very small pieces of caudal membrane at posterior tip. A regenerate of 11 days in straight Ringer solution.

In each of the above figures an arrow indicates the transition zone between old and new tissues.

bud is of interest, too, because of the fact that there are, at the posterior tip, two very small, thin pieces of tissue which resemble the caudal membrane.

In a few instances tadpoles have lived and regenerated in 1.1 Ringer solution. In each of these cases the bud formed is without the caudal membrane.

In this connection it is interesting to note the report of Avel (1932) that, with the approach of metamorphosis, ability to regenerate becomes less and then disappears in the caudal membrane while at corresponding levels it persists for a long time in the musculo-skeletal axis. No ex-

periments have been carried out to determine whether or not the isotonic solution affects, in a similar manner, all stages in the development of tadpoles. In the present work, however, all control individuals produced well-formed tails with complete caudal membranes. Thus it is quite clear that the results here described are due to the effect of the Ringer solution and not to the approach of metamorphosis alone.

#### SUMMARY

Tadpoles of *Rana palustris*, regenerating new tails, produce, in general, less new tissue when kept in an approximately isotonic frog Ringer solution than when allowed to remain in ordinary fresh water.

The regeneration of the caudal membrane is frequently affected by isotonic solutions. Occasionally the bud of new tissue consists merely of the main axis without any membrane. In some other cases a much reduced or irregular membrane is formed.

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